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HISTOMORPHOLOGICAL CHANGES IN THE TESTES OF *OREOCHROMIS NILOTICUS* DURING BREEDING AND NON BREEDING SEASONS

(With 4 Tables and 10 Figures)

By

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التغيرات الهستومورفولوجية لخصي البلطي النيلي أثناء موسم التكاثر وموسم عدم التكاثر

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اجريت هذة الدراسة على ٥٨ من ذكور اسماك البلطى النيلي لدراسة العلاقة بين الشكل الخارجي والتغيرات الهستومورفولوجية للخصي أثناء موسم التكاثر وموسم عدم التكاثر. أتضح من هذة الدراسة أن ذكور اسماك البلطي النيلي اثناء موسم التكاثر تميزت بلون اسود لامع في المناطق الظهرية والجانبية من الجسم وبلون احمر على المنطقة البطنية والرأس. وقد ظهرت الخصيتان طويلتان، رفيعتان في نفس الحجم تقريبا، وقعتا في الجزء الخلفي من التجويف البطنى أسفل المثانة الهوائية ويرتبطان بجدار الجسم العلوي بمساريقا الخصي. أثناء موسم عدم التكاثر، ظهرت الخصيتان صغيرتان خيطيتان الشكل بلون أبيض مع القليل من الأوعية الدموية، حيث كان متوسط طول الخصية ٣.٩٦ ± ١١. سم، وقطرها ١٣, ٣ ± ٠.٢٣مم، ومتوسط وزنها ٦.٣٣± ٠.٨ جم بينما أثناء موسم التكاثر، ظهرت الخصيتان بلون وردى مع الزيادة في الوزن وظهرت الأوعية الدموية واضحة وكذلك خرج المنى بضغط بسيط على الخصية ، وكان متوسط طول الخصية ٤,٤٩ ± ٢, سم، وقطرها٤,٤٦ ±٢, سم، ومتوسط وزنها ١٠,٧٩ ± ٨, جم. لوحظ من خلال الدراسة النسيجية أن خصية البلطي النيلي من النوع الشعاعى حيث تتكون من أنيبيات منوية ملفوفة ومتفرعة، وتبطن هذه الانيبيات بالخلايا المنوية وخلايا سرتولى. وتكونت الانيبيات المنوية من الأكياس الجرثومية، حيث تكون الخلايا الجرثومية الموجودة في الكيس الواحد في نفس المرحلة من النمو. تحاط الخصية بمحفظة تحتوى على عدد قليل من الألياف البيضاء والمرنة وبعض العضلات الملساء، ويزداد سمك المحفظة أثناء موسم عدم التكاثر. أثناء عملية تكوين الطلائع المنوية، تتكاثر خلايا أمهات المنى الأولية لتكون مجموعات من أمهات المنى الثانوية التي تنقسم ميتوزيا لتكون خلايا منوية أولية ، الأخيرة تخضع للانقسامات الميوزية لتعطى خلايا منوية ثانوية والتي تمر بانقسام ميوزى ثاني لتكون العديد من الطلائع المنوية وأثناء التخليق المنوى، تتحور الطلائع المنوية إلى حيوانات منوية ناضجة ويتم ذلك فى تجويف الانبيبات المنوية. أما خلايا سرتولى فقد

ظهرت بشكل كمثرى تحتوى على سيتوبلازم باهت ونواة قاعدية، حيث تكونت جدر الأكياس الجرثومية من بروزات هذه الخلايا. ويزداد عدد خلايا سرتولى. أثناء موسم التكاثر. فيما يتعلق بالخلايا البينية فقد ظهرت في مجموعات في النسيج البيني بين الانبيبات المنوية أثناء موسم التكاثر، تميزت الانبيبات المنوية بوجود المراحل الأخيرة من عملية تكوين الطلائع المنوية والمراحل الأولى من التخليق المنوي. وأثناء موسم عدم التكاثر، تميزت الانبيبات المنوية بوجود المراحل الأولى من عملية تكوين الطلائع المنوية. وانخفضت نسبة أحجام الانبيبات المنوية بينما زادت نسبة أحجام الأنسجة البينية.

SUMMARY

The present work was conducted to highlight the relationship between the external morphology of *O. niloticus* and the histomorphological changes of the testes during the breeding and the non breeding seasons. A total of 58 male fishes of *O. niloticus* were used in this investigation. The males showed bright black coloration on the dorsal and lateral parts of their body and red colour on the ventral part of their body and head during the breeding season. Testes were paired long narrow structure of approximate equal size, locating in the posterior body cavity, attached to the dorsal body wall by mesorchium. During the non-breeding season, the testes were small, thread- like and dull white in colour. During the breeding season, testes were pinkish in colour and increased in weight. The testis was covered with a capsule consisting of few collagenous, elastic fibers. Testicular parenchyma consisted of branched seminiferous tubules and interstitial tissue. The seminiferous tubules were lined with spermatogenic and Sertoli cells. Seminiferous tubules were made up of spermatocysts. During spermatogenesis, primary spermatogonia proliferated to form secondary spermatogonia which divided mitotically to form primary spermatocytes. The later underwent meiotic divisions to form secondary spermatocytes that passed with second meiotic divisions giving many spermatids which transformed into spermatozoa in the lumen of the seminiferous tubules. Sertoli cells were pyriform cells with slightly eosinophilic cytoplasm and one basal nucleus. During the non-breeding seasons, the diameter of the seminiferous tubules reached 102.94 $\pm$ 1..83 µm and the mean number of Sertoli cells was 1.79 $\pm$ 0.16 /spermatocyst. During the breeding season, the diameter of the seminiferous tubules was 124.78 $\pm$ 2.32 µm and the mean number of Sertoli cells was 3.01 $\pm$ 0.14/ spermatocyst.

Key words: *O. niloticus**, testes, breeding***

INTRODUCTION

Nile tilapia belongs to genus Oreochromis. This species is naturally distributed in Palestine, the Nile River as well as most parts of African Rivers & lakes (Trewawas, 1982 and Beamish; Booth and Deacon, 2005). *O. niloticus* is gonochoristic, which each individual possessing a single sexual phenotype. Nile tilapia is characterized by extended spawning seasons, maturity at small size and a fast growth rate. It has been termed the aquatic chicken for its extraordinary production capabilities (Peterson; Slack; Brown- Peterson and McDonald, 2004). Tilapia have one pair of bilateral gonads locating in the posterior part of the body cavity immediately ventral to the swim bladder and attached by mesentries to the parietal peritoneum. Short ducts extend from the posterior end of the gonads to the genital pore. In addition to production of gametes, the gonads also produce hormones from endocrine tissue (Bond, 1979).

The aim of the present investigation is to highlight the relationship between the external morphology of the fish and the histomorphological changes of the gonadal tissue during different seasons of the year. Also, to detect the relationship between some environmental conditions such as water temperature and spawning activity.

MATERIALS and METHODS

The materials employed in this study consisted of randomly obtained 58 male adult specimens *Oreochromis niloticus*. The materials were collected every month from the Nile River at Elkhazan bridge in Assuit city during the year. The specimens ranging from 14.46 $\pm$ 0.22 &13.92 $\pm$ 0.16 cm in standard length and from 96.39 $\pm$ 2.19 g. in body weight. After recording the above mentioned measurements, the fish were dissected as soon as possible to obtain the testes.

***Ttesticular measurements*:** The testes length (cm) was measured individually (right&left) using a ruler from the anterior to the posterior end. In addition their diameter (mm) was measured individually (right&left) using a caliber. Testes (right&left) were also weighed individually using Berekel balance.

Gonadosomatic index (GSI): Monthly variation of gonadosomatic index provides good indication of the extent of development of gonad with respect to the time of year (Hatikakoty and Biswas, 2004).

GSI was calculated monthly from each male fish (Table 1) using the following formula:

GSI % = Gonads (testes) weight (g.) / Body weight (g.) x 100

A regular record of water temperature for every month was recorded for three times using water thermometer and the mean values were taken in order to study the possible correlation between the temperature and the spawning activity of *O. niloticus* (Table 2).

Histological preparation: The samples for histological examination were dissected as soon as possible from the anterior, middle and posterior parts of each testes (1x1x.05 cm) and were immediately fixed in Bouin's fluid for 24 hours. The fixed materials were dehydrated in an ascending series of ethanol, cleared in methyl benzoate and then embedded in paraffin wax. Transverse and longitudinal paraffin sections at 5-8 µm in thickness were cut and stained with the following histological stains; Harris haematoxylin and Eosin (Harri's, 1900), Grossmon's Trichrome (Grossmon, 1937), Periodic Acid –Schiff (PAS) (McManus, 1946), Verhoeff's stain (Verhoeff's, 1908).

Morphometrical measurements: By using Image analysis system (Leica Q500 MC) morphometric study applied on the representative stained sections, including**:** the thickness of tunica albuginea, diameter of seminiferous tubules.The volume percentage of seminiferous tubules and interstitial tssue. Number of Sertoli cells/ cross section of seminiferous tubules as well as per spermatocysts. The data was statistically analyzed by computer program SPSS ver II (Tables 3&4).

RESULTS

The present study showed that during breeding season, both upper & lower jaws of sexually mature males were enlarged making their profile concave. Males exhibited bright gray color, as compared with those examined during non-breeding season. This bright color is lost as soon as this season is over .Breeding males had a distinct black lines appeared on the dorsal and lateral parts of their body, while red colour was found on the ventrolateral parts of their body and head (Fig. 1). These charactrestics are not appeared in non breeding males (Fig. 2).

Testes of male *O. niloticus* were paired long, narrow structure and approximate of equal size. They were located in the posterior part of the body cavity. During the non-breeding season, the testes were small,

flaccid, thread- like and dull white in colour with slight vascularization (Fig. 3). During the breeding season, testes were soft, creamy in texture, turgid, pinkish in colour and increased in weight. The blood vessels were prominent and the milt run with slight pressure (Fig. 4). The mean testicular diameter of mature males used in present investigation during the breeding season was 4.46 $\pm$ 0.20 mm and during the non- breeding season was 3.13 $\pm$ 0.23 mm. The histological investigation showed that the testis was bean- shaped in cross section and cylindrical in longitudinal section. Testis of tilapia was of the radial type, a system of seminiferous tubules passed from the dorsal and lateral wall of the testis to the central lumen (Fig. 5). The central lumen led to efferent ducts, which opened into the sperm duct or vas deferens that led to the urogenital pore (Fig. 6). The testis was enclosed completely by tunica albuginea, which was consisted of few collagenous fibers, some smooth muscle cells and elastic fibers. The testicular parenchyma was consisted of branching tubular seminiferous tubules & interstitial tissue; a)- *The seminiferous tubules;* The seminiferous tubules was delimited by a basal lamina that showed PAS positive reaction and had a central lumen surrounded by the germinal epithelium which was made up of spermatogenic cells as well as Sertoli cells. The seminiferous tubules were made up of spermatocysts, that it formed by the cytoplasmic projections of Sertoli cells. The spermatocysts contained spermatogonia, primary spermatocytes, secondary spermatocytes and spermatids, while spermatozoa found in the lumen of the seminiferous tubule. Within each spermatocyst, the germ cells were at the same stage of development (Fig. 7).

Spermatogenesis:

The process of sperm formation occurred in spermatocysts and consisted of two distinct phases:

1 - Renewal, mitotic proliferation of spermatogonia.

2 - Meiosis followed by spermiogenesis.

The primary spermatogonia (sperm mother cells) were the largest cells of the germinative lineage, that could occur either isolated or in groups inside the cysts in the seminiferous tubules. They were large oval cells with very scarce, lightly basophilic cytoplasm and large round nucleus with a single nucleolus. Primary spermatogonia were found during the breeding and non-breeding seasons and were usually present within those seminiferous tubules locating peripherally under the tunica albuginea (Fig. 8). Primary spermatogonia proliferate to form clusters of secondary spermatogonia, which were rounded cells, smaller than

primary spermatogonia. Each cluster was enclosed in a cyst and divided synchronously, mitotically to form primary spermatocytes that were smaller in size than secondary spermatogonia, the nucleus was strongly stained with heamatoxylin and the cytoplasm had little affinity for dyes. Meiotic divisions of primary spermatocytes produced secondary spermatocytes that were somewhat smaller than primary spermatocytes with weakly stained nucleus Second meiotic division produced haploid spermatids, those were small with scant cytoplasm, strongly basophilic nucleus. Spermatids inside the cyst increased in number towards the center of the testis. Metamorphosis of spermatids to spermatozoa occurred in the lumen of the testis tubules after the cyst had burst and this process is called spermiogenesis.

Spermatozoa were smaller than spermatids with strongly basophilic nucleus. They were found in the interior of the seminiferous tubules (Figs. 7), efferent ducts and sperm duct. Sertoli cells: Pyriform cells with slightly eosinophilic cytoplasm and one basal clear nucleus had an irregular contour with one clear nucleolus. Their adluminar surface was thrown into finger- like projections, whereas their basal surface was thrown into a complex system of stubby projections (Fig. 9). Their projections formed the borders of spermatocysts. Spermatogonia and spermatocytes did not touch the basal lamina, they were surrounded and enclosed by cytoplasmic expansions of Sertoli cells. Spermatids and earlier stages of spermatogenesis were associated with Sertoli cells (Fig. 10). b)- *Interstitial tissue:* The interstitial tissues occured between the seminiferous tubules, contained interstitial cells, fibroblasts, blood vessels and some collagenous fibers. The mean volume percentage of interstitial tissue during the non-breeding season was 30.8 $\pm$ 1.72 %, while during the breeding season was 21.54 $\pm$ 0.81 %. *Interstitial cells (Leydig cells):* Occupied greater part of the testis interstitum. They were formed of small or large clusters of polygonal cells containing an ovoid nucleus. These cells were located in the fibrous supporting connective tissue between seminiferous tubules (Fig. 10). During the non breeding season the tunica albugeia was thick and contained large amount of elastic fibers and smooth muscle fibers. The volume percentage of seminiferous tubules was reduced, and its mean value reached 70.19 $\pm$ 1.0 %. The minimum mean value was 68.23 $\pm$ 0.87 % in February (Table 3). The diameter of the seminiferous tubules reduced, and its mean diameter reached 102.94 $\pm$ 1.83 µm. The minimum mean value was 96.85 $\pm$ 0.87 µm in February. Seminiferous tubules were dominated by early stages of spermatogenesis (spermatogonia and primary

spermatocytes) and had a narrow lumen with considerably fewer spermatocysts. In addition during the non-breeding season, the mean number of Sertoli cells was 6.4 ± 0.5 / cross section of seminiferous tubules and 1.79 ± 0.16 / spermatocyst. The minimum number was 5 ± 0.31 / cross section of seminiferous tubules in February and 1.33 ± 0.21 / spermatocyst in February (Table 4). The volume percentage of interstitial tissue increased during the non-breeding season, and its mean value reached 29.8 ± 1.72 %. The maximum value was 36.77 ± 1.0 % in February (Table 3). During the breeding season the tunica albuginea was thin during the breeding season its mean thickness reached 7.10 ± 0.36 µm and the minimum mean value was 6.35 ± 0.69 µm in April. The volume percentage of seminiferous tubules increased, and its mean value reached 78.43 ± 0.81 %. The maximum mean value was 80.11 ± 0.96 % in June.The diameter of the seminiferous tubules was significantly increased during the breeding season, and its mean diameter reached 124.78 ± 2.32 µm. The maximum mean value was 141.51 ± 1.21 µm in July (Table 3). The seminiferous tubules widened and elongated toward the center of the testis where their lumen showed abundant sperms. The sperms were free and were almost neither linked nor bound to Sertoli cells. The seminiferous tubules were dominated by later stages of spermatogenesis (secondary spermatocytes and spermatids) and early stage of spermiogenesis (spermatids). Seminiferous tubules were strongly packed with spermatozoa in pro-spawning and spawning periods. During the breeding season, there was a significant increase in the number of Sertoli cells, its mean number reached 10.6 ± 0.67 / cross section of seminiferous tubules and 3.01 ± 0.14 / spermatocyst. The maximum mean number was 13 ± 1.37 /cross section of seminiferous tubules in May and 3.5 ± 0.42 / spermatocyst in June (Table 4). The volume percentage of interstitial tissue decreased during the breeding season, and its mean value reached 21.54 ± 0.81 % (Table 4).

Gonadosomatic index:

GSI= gonads weight \ body weight x 100

GSI can be used as indicator for gonadal development. When gonadosomatic index reach a maximum value, this gives a perfect indication to the time of spawning.

Gonadosomatic indix were highest between May to July. GSI peaked in July (Table 1). The mean GSI during the breeding season was 11.15 ± 0.39 % While the mean GSI during the non- breeding season was 7.91 ± 0.39 % (Table 1). From the present investigation, the water

temperatures were highly correlated with GSI and the optimum temperature for spawning was between 21°C to 24°C (from May to July) that was the peak period for the GSI (Table 2).

Table 1: Average monthly fluctuations in GSI.

	Number of specimens	Mean body weight (gm)	Mean testis weight (gm)	GSI
Non-breeding season				
October	5	97.2	8.3	8.53
November	5	95.0	7.6	8.0
December	5	90.0	6.6	7.33
January	5	82.4	6.0	7.28
February	5	83.2	5.9	7.09
March	4	96.25	8.9	9.24
M±SE			7.0	7.91
breeding season	---------	-------	--------	-------
April	5	106.2	11.2	10.54
May	5	103.2	11.4	11.04
June	5	104.2	12.7	12.18
July	5	103.0	12.8	12.42
August	4	100.75	10.75	10.66
September	5	99.1	10.1	10.1
M±SE			11.77	11.5

Table 2: The mean values of water temperature during different months of the year.

Month	Temperature
October	18 °C
November	16 °C
December	11 °C
January	11 °C
February	13 °C
March	15 °C
April	19 °C
May	21 °C
June	22 °C
July	24 °C
August	25 °C
September	26 °C

Table 3: The mean volume percentage of seminiferous tubules, interstitial tissue and diameter of seminiferous tubules (µm) during different months of year.

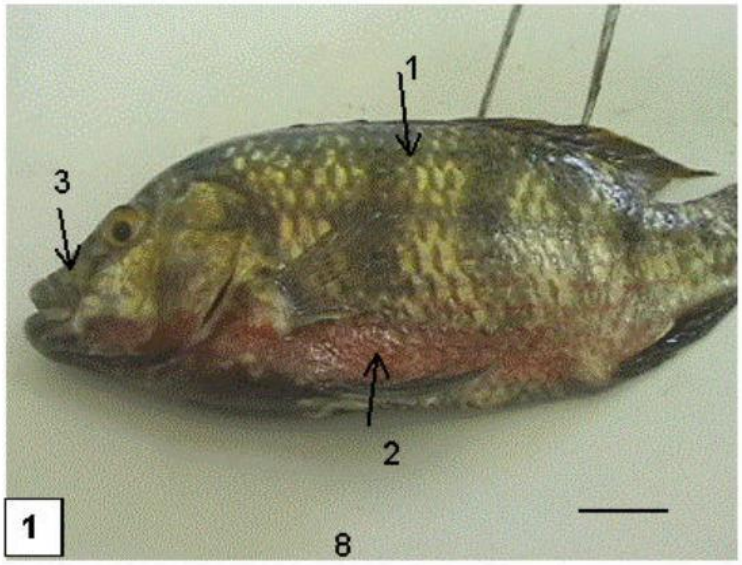
Months	Volume percentage of seminiferous tubules (%)	Volume percentage of interstitial tissue (%)	Diameter of the seminiferous tubules (µm)
Non-breeding season October	69.3 ± 0.97	30.8 ± 0.88	106.28 ± 1.66
November	69.36 ± 1.78	30.64 ± 1.88	101.26 ± 2.0
December	70.27 ± 0.99	29.73± 0.97	102.37 ± 1.85
January	69.12 ± 1.08	30.88 ± 2.76	99.21 ± 1.11
February	68.23 ± 0.87	36.77 ±1.00	96.85 ± 0.87
March	74.00 ± 2.00	26.00 ± 0.96	109.0 ± 0.95
M±SE	70.19 ± 1.0	29.8 ± 1.72	102.94 ± 1.83
Breeding season April	78.75 ± 1.23	21.25± 1.44	112.1 ± 0.93
May	79.11 ± 0.87	20.78 ± 2.38	119.52 ± 1.24
June	80.11 ±0.96	19.89 ± 0.64	132.11 + 1.98
July	78.88 ± 1.09	21.12 ± 1.04	141.51 ± 1.21
August	76.33 ± 0.95	24.67± 1.55	125.0 ± 1.22
September	76.0 ± 0.78	24.0 ± 0.88	118.44 ± 0.83
M±SE	78.43 ± 0.81	21.54 ± 0.81	124.78 ± 2.32 *

Values were represented by mean ± standard error (SE).
* means significant.

Table 4: The mean thickness of the tunica albuginea, number of Sertoli cells / cross section of seminiferous tubules and / spermatocyst during different months of the year.

Months	Thickness of tunica albuginea (µm)	Number of Sertoli' cells/ cross section	Number of Sertoli' cells/ spermatocyst
Non-breeding season October	14.35 ± 0.36	6.5 ± 0.85	1.8 ± 0.66
November	10.98 ± 0.78	7 ± 0.44	1.83 ± 0.3
December	14.02 ± o.75	6 ± 0.70	1.83 ± 0.16
January	14.38 ± 0.88	6 ± 0.70	1.66 ± 0.33
February	16.70 ± 1.76	5 ± 0.31	1.33 ± 0.21
March	15.58 ± 1.86	8 ± 0.83	2.33 ± 0.49
M±SE	14.33 ± 0.96	6.4 ± 0.5	1.79 ± 0.16
Breeding season April	6.35 ± 0.69	9 ± 1.30	2.83 ± 0.30
May	6.55 ± 0.56	13 ± 1.37	3.0 ± 0.44
June	6.80 + 1.06	10 ± 0.70	3.5 ± 0.42
July	7.48 ± 0.51	11 ± 0.44	3.1 ± 0.16
August	8.33 ± 0.71	10 ± 0.70	2.66 ± 0.33
Sebtember	7.0 ± 0.74	10.5 ± 0.33	3.2 ± 0.11
M±SE	7.1 ± 0.36 **	10.6 ± 0.67 n.s.	3.01 ± 0.14 **

Values were represented by mean ± standard error (SE).
n.s. means not significant. ** means highly significant.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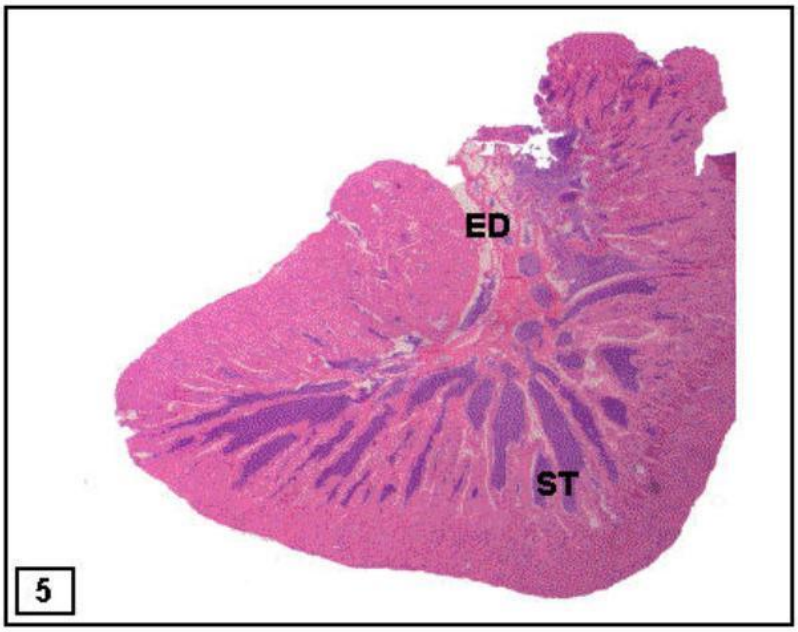
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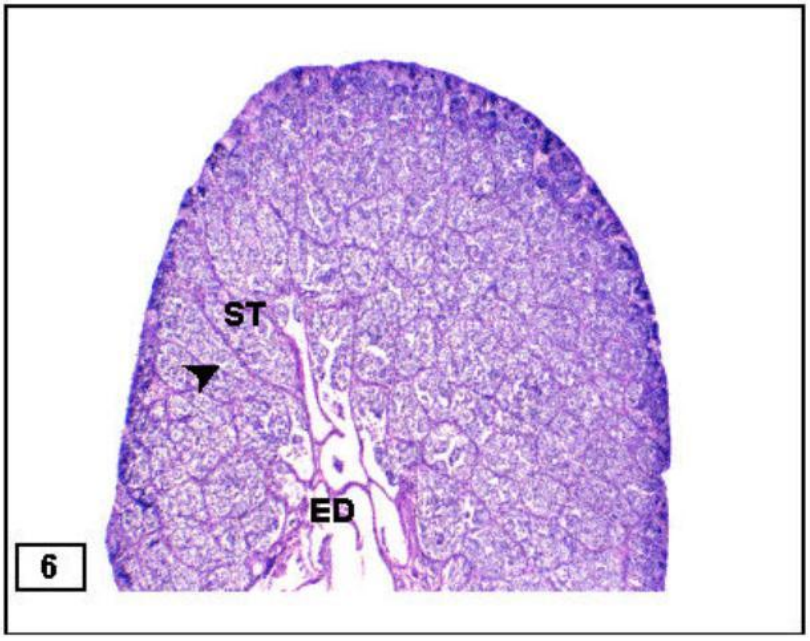
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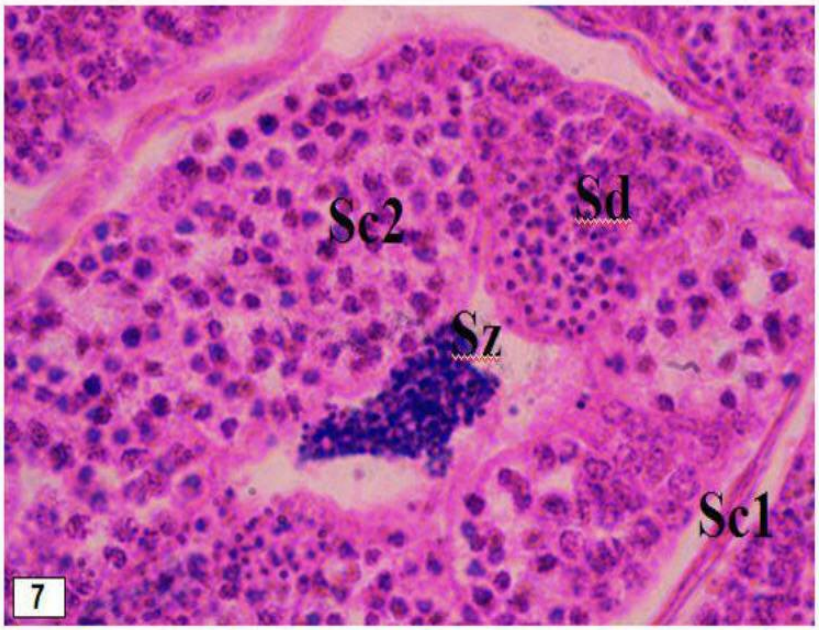
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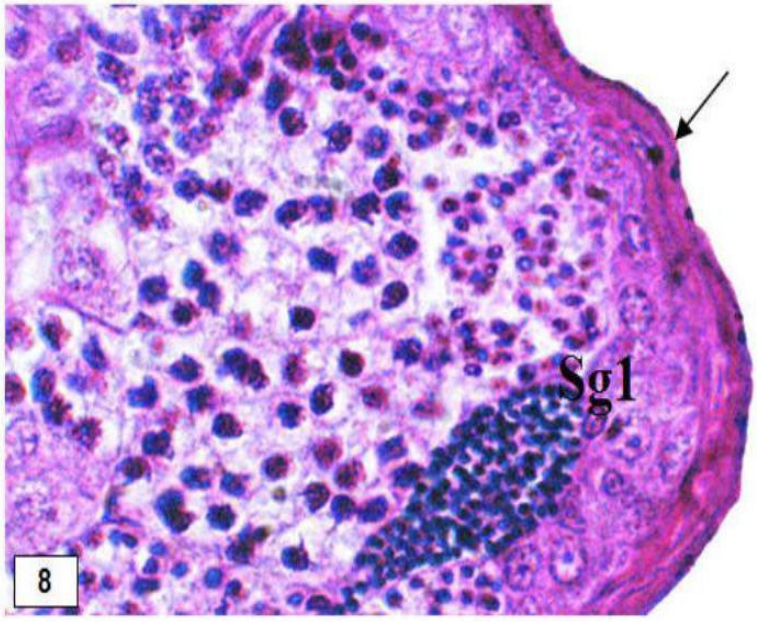
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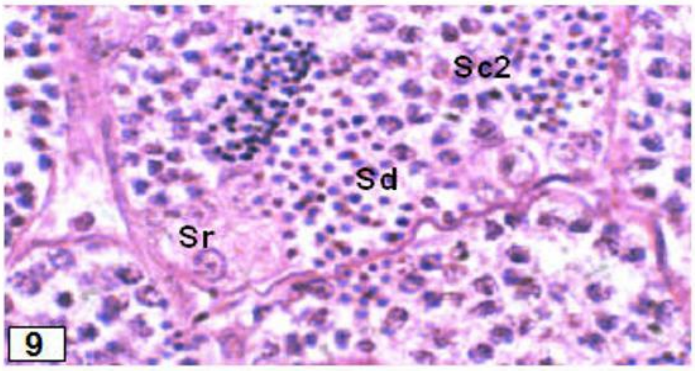

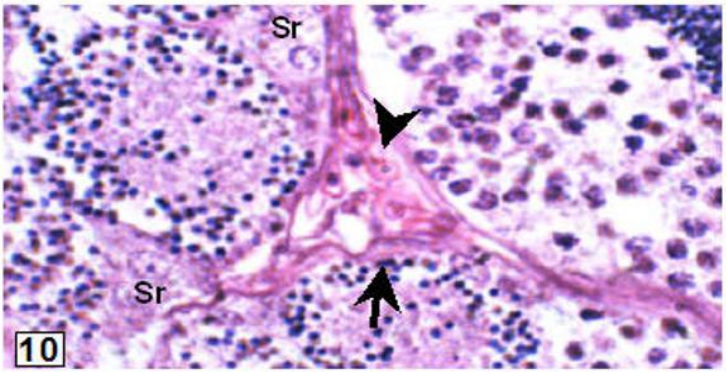

LEGENDS

Fig. 1: Lateral view of the male *O. niloticus* during the breeding season, showing a distinct bright black color on the dorsal and lateral areas of the body (arrow 1), red color on the ventrolateral parts of their body and head (arrow 2) and the jaws enlarged making their profile concave (arrow 3). (Bar = 0.85)

Fig. 2: Lateral view of the male *O. niloticus* during the non-breeding season, showing the absence of the bright black and red color and the jaws reduced in size making their profile convex (arrow). (Bar= 0.85)

Fig. 3: Photograph of the testes of O. niloticus during the non-breeding season. The testes were small dull white in color.

Fig. 4: Photograph of the testes of O. niloticus during the breeding season. The testes were large white in color.

Fig. 5: Cross section in the testis showing distended seminiferous tubules (ST) that opened in the center of the testis in the efferent ducts (ED).(Heamatoxylin and Eosin, X 10)

Fig. 6: Photomicrograph of the testis showing seminiferous tubules (ST) that opened in the center of the testis in the efferent ducts (ED).(Periodic acid Schiff reagent, X 50)

Fig. 7: Photomicrograph in the testis during the breeding season showing seminiferous tubule contained primary spermatocytes (Sc1), secondary spermatocytes (Sc2), spermatids (Sd) and spermatozoa (Sz). (Heamatoxylin and Eosin, X 400)

Fig. 8: Photomicrograph in the testis showing the thick tunica albuginea (arrow). Notice the presence of primary spermatogonia (Sg1) under the tunica albuginea. (Heamatoxylin and Eosin, X 400

Fig. 9: Photomicrograph in the testis showing seminiferous tubule contained secondary spermatocytes (Sc2), spermatid (Sd) and Sertoli cells (Sr). (Heamatoxylin and Eosin, X 400)

Fig. 10: Photomicrograph in the testis showing interstitial tissue (arrow) contained interstitial cells (arrow head). Notice the presence of Sertoli cells (Sr). (Heamatoxylin and Eosin, X 1000)

DISCUSSION

The present work was carried out on 58 specimens of male*O. niloticus* throughout the year, in order to observe the morphological and histological changes in the testes during different seasons of the year. The result showed that the breeding season for reproduction was between April and September, while non-breeding season was between October and March, the current findings simulate those of Caputo, V.; Mesa, M.L.; Candi, G. and Cerioni, P.N. (2003) and Cinquetti and Dramis (2003). The testicular morphology observed in the present study is similar to that described in the catfish Arockiaraj, A.J.; Haniffa, M.A.; Seetharaman, S. and Singh, S. (2004) and *Oreochromis mossambicus*

(Hatikakoty and Biswas, 2004). As testes develop, they present accentuated differences in size and form. The mature stage was well evidenced by its largest volume corresponding to increasing size of the seminiferous tubules and quantity of spermatozoa. Variation in the form occured from the thread- like appearance during the non-breeding season and becoming large and cylindrical during the breeding season, and resulting wrinkled after spawning. The present investigation revealed that the testes were covered by the tunica albuginea that formed of collagenous and elastic fibers, fibroblasts and smooth muscle fibers. The tunica albuginea was thin during the breeding season as a result of progressive expansion of the testis by maturation, its mean thickness reached 7.10 $\pm$ 0.36 µm. while during the non-breeding season, its mean thickness reached 14.33 $\pm$ 0.96 µm. The elastic fibers that found in the wall and especially visible after spawning are responsible for contraction of the testis and discharge of sperms, similar observation was reported by (Arockiaraj, *et al*., 2004). The current observations revealed the presence of large amounts of spermatozoa in the lumen of the distended seminiferous tubules and in the sperm ducts leading to increase in weight of the testes to reach its maximum weight during the breeding season (11.77 $\pm$ 0.41 gm). On the other hand, the testes during the non-breeding season appeared flaccid and dull white in coloure and its mean weight was 7.0 $\pm$ 0.56 gm.Testis of tilapia is of radial type where seminiferous tubules arranged radially from testis periphery toward the sperm duct. The sperm duct results from connection of efferent ducts at the center of the testis and this applied with Alka'abi (1996). Testis of *O. niloticus* was composed of coiled tubular branching seminiferous tubules that were delimited by a basal lamina, and contained both germ and Sertoli cells. Somatic cells, such as Leydig cells and blood vessels, were located between the tubules. Seminiferous tubules had a central lumen surrounded by the germinal epithelium. This was organized in spermatocysts, where spermatogenesis occurred. The walls of spermatocysts were formed by cytoplasmic processes of Sertoli cells. Each cyst contained germ cells at the same stage of development, these are similar to the observations of Schulz, R.W.; Menting, S.; Bogerd, J.; Franca, L.R.; Vilela, D.A. and Godinho, H.P. (2005). The testicular organization of *O. niloticus* corresponds to the unrestricted (cystic) type, where spermatogenesis is completed within spermatocysts and leads to synchronous development of germ cells. These results agreed with Mattei, X.; Siau, Y.; Thiaw, O.T. and Thiam, D. (1993) and Hamdoon and Zayed (1998). Spermatogenesis occured in a series of a process,

which involves; a) proliferation of spermatogonia through repeated mitotic divisions; b) growth of spermatogonia to form primary spermatocytes; c) then undergo reduction division to form secondary spermatocytes; d) the division of secondary spermatocytes produces spermatids; and e) the metamorphosis occured to form the motile spermatozoa. Mature spermatocysts open to release cohorts of sperm into a central lumen, which lead to a contagious system of efferent ducts. Similar findings were reported by Amiri, *et al*. (1996) who studied the testicular development and serum sex steroid profiles during the annual sexual cycle of the male sturgeon hybrid**;** Redding and Patino (1993) who studied the reproductive physiology of fishes and Gomes and Araujo (2004) who studied the reproductive biology of two marine catfishes. The present study revealed significantly increase in number of Sertoli cells per spermatocyst during the breeding season, such observation is similar to that mentioned by Sculz, *et al.* (2005), however, evidence for proliferation such an increase mitotic divisions in Sertoli cells was not observed. In addition, it can be suggested that, during the non-breeding season, the Sertoli cells regress or severally reduced in size that could not be visible by the light microscopy, however during the spawning or breeding season, these cells flourished up and increased in size that made it microscopically visible. Moreover, further investigations are required to identify the possible reasons of increased number of Sertoli cells per spermatocyst during breeding season in the Nile Tilapia.In the same respect, Hibiya (1982) believed that Sertoli cells play a role in supplying of nutrients to the germ cells, while Redding and Patino (1993) mentioned that these cells are responsible for the physical support and regulation of spermatogenesis. On the other hand, Cinquetti and Dramis (2003) stated that Sertoli cells were involved in phagocytosis of degenerating, residual sperm cells and in spermatogonial proliferation. In addition, these cells considered to be the main constituent of the blood testicular barrier, which developes usually before or after meiosis and it maintains features of intense protein synthesis throughout the whole spawning season. In agreement with Redding and Patino (1993), Leydig (interstitial, interlobular) cells are interspersed in the connective tissue surrounding the seminiferous tubules; their primary function is to produce sex steroids (testosterone) needed for gametogenesis and expression of secondary sex characteristics. From observations of the present study, it was found that during spermiogenesis, the spermatids mature and develop to become spermatozoa, spermatozoa is collected in the efferent tubules, from

which they are expelled during spermiation (sperm release). The efferent ducts of the testes join to form spermatic ducts. The paired spermatic ducts coalesce distally to form the sperm duct which emptied into urogenital. These findings in agreement with the results obtained by Amiri, *et al*. (1996).

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